Citation:

Reaven GM, Abbasi F, Bernhart S, Coulston A, Darnell B, Dashti N, Kim H, Kulkarni K, Lamendola C, McLaughlin T, Osterlund L, Schaff P, Segrest J. Insulin resistance, dietary cholesterol and cholesterol concentration in postmenopausal women. *Metabolism.* 2001 May; 50 (5): 594-597.

PubMed ID: 11319723

Study Design:

Randomized Controlled Trial

Class:

A - Click here for explanation of classification scheme.

Research Design and Implementation Rating:

NEUTRAL: See Research Design and Implementation Criteria Checklist below.

Research Purpose:

To compare various aspects of glucose, insulin and lipoprotein metabolism before and three months after increases in daily dietary cholesterol intake from 113mg to either 319mg, 523mg or 941mg in postmenopausal women, stratified into an insulin-sensitive and insulin-resistant group.

Inclusion Criteria:

- Non-diabetic women who were postmenopausal for at least one year
- Body mass index (BMI) between 19 and 33kg/m^2
- A fasting plasma total cholesterol concentration <280mg/dL
- A triglyceride concentration <400mg/dL
- Normal results from a physical examination, hemogram and routine biochemical tests
- Subject using hormone replacement therapy were included, other medications or dietary supplements were included, but continued on them throughout the study.

Exclusion Criteria:

Women with steady state plasma glucose (SSPG) levels not <100mg/dL or >160 mg/dL.

Description of Study Protocol:

Recruitment

Not described.

Design

- Randomized controlled trial
- Subjects were studied over a 12-week period, four weeks on the baseline diet, a four-week washout period, followed by a second four-week diet period consuming varied levels of dietary cholesterol.

Dietary Intake/Dietary Assessment Methodology

- Not applicable (Subjects were provided with all foods consumed as part of the intervention)
- No methodology to assess compliance was reported.

Blinding Used

Subjects were blinded to which cholesterol level they received during the intervention.

Intervention

- Subjects were started on a baseline, low-cholesterol diet that contained 113mg of cholesterol per day
- Subjects were then randomized to one of three experimental diets that contained either 319mg, 523mg or 941mg per day of cholesterol
- Various combinations of egg and egg substitute were used to attain the desired amount of cholesterol in each of the four diets
- Caloric level was determined for each subject using the Harris-Benedict equation, and diets were designed to maintain body weight within 0.5kg of baseline weight throughout the study
- Subjects were required to visit the research center daily to check body weight, eat one meal (that contained all of the dietary cholesterol for the day) and pick up meals
- Each of the four diets conformed to the NCEP Step 1 diet, which was 20% protein, 50% carbohydrate, 30% fat, 9% saturated fat, 9% polyunsaturated fat and 12% monounsaturated fat
- Each of the four diets was identical in terms of macro- and micro-nutrients, such that only cholesterol content varied.

Statistical Analysis

Student's paired T-test was performed the evaluate the effect of cholesterol intake on plasma cholesterol concentration.

Data Collection Summary:

Timing of Measurements

- Subjects were studied over a 12-week period, four weeks on the baseline diet, a four-week washout period, followed by a second four-week diet period consuming varied levels of dietary cholesterol
- At the end of each four-week diet period, subjects were readmitted to clinic for metabolic measurements.

Dependent Variables

Blood cholesterol concentrations (total, LDL, real LDL, HDL, HDL_2, HDL_3, LDL/HDL, IDL, VLDL, $VLDL_3$), triglycerides, Apo A-1, Apo A-2, Apo B, Apo C-3, and Apo E were all measured after an overnightfast.

Independent Variables

Dietary intake of cholesterol.

Control Variables

SSPG status.

Description of Actual Data Sample:

- *Initial N:* Not reported
- *Attrition (final N):* N=65
  - 32 insulin-sensitive women
  - 33 insulin-resistant women
- *Age:*
  - 319mg/dL group: 56±1 years
  - 523mg/dL group: 54±1 years
  - 941mg/dL group: 57±1 years
  - Insulin-sensitive group: 57±1 years
  - Insulin-resistant group: 55±1 years
- *Ethnicity:* Not reported
- *Other relevant demographics:* None reported
- *Anthropometrics:*
  - 319mg/dL group: 26.1±0.9kg/m^2
  - 523mg/dL group: 26.8±0.8kg/m^2
  - 941mg/dL group: 25.4±0.8kg/m^2
  - Insulin-sensitive group: 23.8±0.8kg/m^2
  - Insulin-resistant group: 28.1±0.6kg/m^2
- *Other:*
  - Baseline characteristics of the three cholesterol groups did not differ on any of the measured variables
  - Insulin-sensitive women had significantly lower BMI, LDL/HDL, triglycerides and Apo-B, and significantly higher HDL compared to insulin-resistant women
- *Location:* United States.

Summary of Results:

Total cholesterol and LDL-cholesterol concentrations changed very little in both the insulin-sensitive and insulin-resistant groups when cholesterol intake was increased.

	Total Cholesterol (mg/dL)		LDL Cholesterol (mg/dL)	
Group	**Baseline**	**After Intervention**	**Baseline**	**After Intervention**
319 mg/dL				
Total (N=23)	167±6	178±6	105±5	112±5

Sensitive (N=12)	168±8	172±2	101±6	105±6
Resistant (N=11)	167±9	184±8	109±7	121±7
523 mg/dL				
Total (N=20)	175±9	181±8	112±7	116±7
Sensitive (N=8)	190±15	196±12	122±13	127±12
Resistance (N=12)	166 ±11	171±10	105±8	108±7
941 mg/dL				
Total (N=22)	176±6	184±6	107±5	113±55
Sensitive (N=11)	167±9	176±9	100±6	107±7
Sensitive (N=11)	184±8	192±9	114±9	120±8

Author Conclusion:

Relatively large increments of dietary cholesterol intake had little effect on total and LDL-cholesterol concentrations in healthy, postmenopausal women, irrespective of whether they were insulin-sensitive or insulin-resistant.

Reviewer Comments:

- *Small sample sizes were used in sub-group analyses*
- *Compliance with the study diet was not reported.*

Research Design and Implementation Criteria Checklist: Primary Research

Relevance Questions

1.	Would implementing the studied intervention or procedure (if found successful) result in improved outcomes for the patients/clients/population group? (Not Applicable for some epidemiological studies)	Yes
2.	Did the authors study an outcome (dependent variable) or topic that the patients/clients/population group would care about?	Yes
3.	Is the focus of the intervention or procedure (independent variable) or topic of study a common issue of concern to nutrition or dietetics practice?	Yes

4.	Is the intervention or procedure feasible? (NA for some epidemiological studies)	Yes

Validity Questions

1.	**Was the research question clearly stated?**	Yes
1.1.	Was (were) the specific intervention(s) or procedure(s) [independent variable(s)] identified?	Yes
1.2.	Was (were) the outcome(s) [dependent variable(s)] clearly indicated?	Yes
1.3.	Were the target population and setting specified?	Yes
2.	**Was the selection of study subjects/patients free from bias?**	Yes
2.1.	Were inclusion/exclusion criteria specified (e.g., risk, point in disease progression, diagnostic or prognosis criteria), and with sufficient detail and without omitting criteria critical to the study?	Yes
2.2.	Were criteria applied equally to all study groups?	Yes
2.3.	Were health, demographics, and other characteristics of subjects described?	Yes
2.4.	Were the subjects/patients a representative sample of the relevant population?	No
3.	**Were study groups comparable?**	Yes
3.1.	Was the method of assigning subjects/patients to groups described and unbiased? (Method of randomization identified if RCT)	No
3.2.	Were distribution of disease status, prognostic factors, and other factors (e.g., demographics) similar across study groups at baseline?	Yes
3.3.	Were concurrent controls used? (Concurrent preferred over historical controls.)	Yes
3.4.	If cohort study or cross-sectional study, were groups comparable on important confounding factors and/or were preexisting differences accounted for by using appropriate adjustments in statistical analysis?	N/A
3.5.	If case control or cross-sectional study, were potential confounding factors comparable for cases and controls? (If case series or trial with subjects serving as own control, this criterion is not applicable. Criterion may not be applicable in some cross-sectional studies.)	N/A
3.6.	If diagnostic test, was there an independent blind comparison with an appropriate reference standard (e.g., "gold standard")?	N/A
4.	**Was method of handling withdrawals described?**	Yes

4.1.	Were follow-up methods described and the same for all groups?	Yes
4.2.	Was the number, characteristics of withdrawals (i.e., dropouts, lost to follow up, attrition rate) and/or response rate (cross-sectional studies) described for each group? (Follow up goal for a strong study is 80%.)	No
4.3.	Were all enrolled subjects/patients (in the original sample) accounted for?	Yes
4.4.	Were reasons for withdrawals similar across groups?	N/A
4.5.	If diagnostic test, was decision to perform reference test not dependent on results of test under study?	N/A
5.	**Was blinding used to prevent introduction of bias?**	Yes
5.1.	In intervention study, were subjects, clinicians/practitioners, and investigators blinded to treatment group, as appropriate?	Yes
5.2.	Were data collectors blinded for outcomes assessment? (If outcome is measured using an objective test, such as a lab value, this criterion is assumed to be met.)	Yes
5.3.	In cohort study or cross-sectional study, were measurements of outcomes and risk factors blinded?	N/A
5.4.	In case control study, was case definition explicit and case ascertainment not influenced by exposure status?	N/A
5.5.	In diagnostic study, were test results blinded to patient history and other test results?	N/A
6.	**Were intervention/therapeutic regimens/exposure factor or procedure and any comparison(s) described in detail? Were interveningfactors described?**	Yes
6.1.	In RCT or other intervention trial, were protocols described for all regimens studied?	Yes
6.2.	In observational study, were interventions, study settings, and clinicians/provider described?	N/A
6.3.	Was the intensity and duration of the intervention or exposure factor sufficient to produce a meaningful effect?	Yes
6.4.	Was the amount of exposure and, if relevant, subject/patient compliance measured?	Yes
6.5.	Were co-interventions (e.g., ancillary treatments, other therapies) described?	N/A
6.6.	Were extra or unplanned treatments described?	N/A
6.7.	Was the information for 6.4, 6.5, and 6.6 assessed the same way for all groups?	Yes
6.8.	In diagnostic study, were details of test administration and replication sufficient?	N/A

7.	**Were outcomes clearly defined and the measurements valid and reliable?**	Yes
7.1.	Were primary and secondary endpoints described and relevant to the question?	Yes
7.2.	Were nutrition measures appropriate to question and outcomes of concern?	N/A
7.3.	Was the period of follow-up long enough for important outcome(s) to occur?	Yes
7.4.	Were the observations and measurements based on standard, valid, and reliable data collection instruments/tests/procedures?	Yes
7.5.	Was the measurement of effect at an appropriate level of precision?	???
7.6.	Were other factors accounted for (measured) that could affect outcomes?	No
7.7.	Were the measurements conducted consistently across groups?	Yes
8.	**Was the statistical analysis appropriate for the study design and type of outcome indicators?**	Yes
8.1.	Were statistical analyses adequately described and the results reported appropriately?	Yes
8.2.	Were correct statistical tests used and assumptions of test not violated?	Yes
8.3.	Were statistics reported with levels of significance and/or confidence intervals?	Yes
8.4.	Was "intent to treat" analysis of outcomes done (and as appropriate, was there an analysis of outcomes for those maximally exposed or a dose-response analysis)?	N/A
8.5.	Were adequate adjustments made for effects of confounding factors that might have affected the outcomes (e.g., multivariate analyses)?	No
8.6.	Was clinical significance as well as statistical significance reported?	Yes
8.7.	If negative findings, was a power calculation reported to address type 2 error?	No
9.	**Are conclusions supported by results with biases and limitations taken into consideration?**	Yes
9.1.	Is there a discussion of findings?	Yes
9.2.	Are biases and study limitations identified and discussed?	No
10.	**Is bias due to study's funding or sponsorship unlikely?**	Yes
10.1.	Were sources of funding and investigators' affiliations described?	Yes
10.2.	Was the study free from apparent conflict of interest?	Yes